Electronic Supplementary information

Exploring the interactions between model proteins and Pd(II) or Pt(II) compounds bearing charged N,N-pyridylbenzimidazole bidentate ligands by X-ray crystallography

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Table S1. Data collection and refinement statistics.

	Lysozyme-**1**	Lysozyme-**2**	Lysozyme-**3**	Lysozyme-**4**	RNase A-**1**
Data Collection					
Diffraction source	Rotating Anode	Rotating Anode	Rotating Anode	Rotating Anode	Rotating Anode
Wavelength (Å)	1.5418	1.5418	1.5418	1.5418	1.5418
Temperature (K)	100	100	100	100	100
Space group	$P4_32_12$	$P4_32_12$	$P4_32_12$	$P4_32_12$	C2
a (Å)	79.487	78.361	78.443	78.108	100.282
b (Å)	79.487	78.361	78.443	78.108	32.805
c (Å)	37.309	36.962	37.385	37.172	72661
β	90.00	90.00	90.00	90.00	90.61
Resolution range (Å)	56.21-1.96 (1.99-1.96)	55.41-2.43 (2.48-2.43)	55.47-1.99 (2.02-1.99)	55.23-2.25 (2.29-2.25)	72.66-2.65 (2.70-2.65)
Total No. of reflections	52429	29365	40090	29555	16853
No. of unique reflections	8938	4616	8433	5705	6649
Completeness (%)	99.3 (98.6)	99.1 (97.0)	99.6 (99.5)	97.6 (95.3)	91.1 (79.2)
$CC_{1/2}$	(0.775)	(0.644)	(0.827)	(0.755)	(0.834)
Multiplicity	5.9 (3.5)	6.4 (4.5)	4.8 (4.0)	5.2 (3.4)	2.5 (2.3)
$\langle I/\sigma(I)\rangle$	15.3 (1.5)	10.4 (1.3)	12.1 (1.7)	9.3 (1.6)	12.1 (1.6)
$R_{merge.}$	0.107 (0.505)	0.174 (0.719)	0.107 (0.569)	0.135 (0.559)	0.090 (0.391)
Refinement					
Resolution range (Å)	56.21-1.96	55.41-2.43	55.47-1.99	55.33-2.25	72.66-2.65
No. of reflections, working set	8480	4378	7994	5396	6196
No. of reflections, test set	434	221	413	293	309
Final R_{cryst} (%)	16.9	20.1	16.6	17.2	18.7
Final R_{free} (%)	23.1	27.3	24.2	27.9	25.5
No. of non-H atoms	1394	1151	1190	1168	2022
R.m.s. deviations from ideal geometry					
Bonds (Å)	0.016	0.012	0.016	0.012	0.012
Angles (°)	2.21	1.82	1.74	1.66	1.65
Average *B* factors (Å^2)	27.26	37.59	25.86	28.70	48.96
Occupancy of Pd/Pt centres	0.65, 0.60,0.60, 0.55, 0.50,0.40, 0.30	0.40, 0.25, 0.25, 0.20	0.70, 0.20	0.45, 0.30	0.70, 0.50, 0.50, 0.20
B-factors of Pt/Pd centres	23.8, 26.7, 31.7, 33.2, 29.4,42.8, 42.9	53.7, 48.6, 46.8, 62.5	34.7, 54.2	50.2, 52.2	33.6, 55.0, 44.0, 52.5
Ramachandran plot‡					
Most favoured (%)	99.0	96.7	97.4	95.7	95.0
Allowed (%)	1.0	3.3	2.6	4.3	4.1
Outliers	0	0	0	0	0.8

Values in parentheses are for the outer shell.

‡Calculated for non-glycine and non-proline residues using Coot.

Table S2. Root mean square deviations (rmsd, Å) among the relative positions of carbon alpha atoms of the lysozyme adduct here solved.

	Lysozyme-**1**	Lysozyme-**2**	Lysozyme-**3**	Lysozyme-**4**	Native lysozyme (pdb code 193L)
Lysozyme-1	-	0.31	0.29	0.32	0.24
Lysozyme-2		-	0.18	0.15	0.25
Lysozyme-3			-	0.16	0.20
Lysozyme-4				-	0.26

Table S3. Structures of protein adducts with Pd compounds reported in the Protein Data Bank.

PDB code	Protein	Pd compound	Resolution (Å)	Pd-protein interaction (< 2.5 Å)	Reference
5YHA	Polyhedra	Pd(allyl) complex (C_3H_5Pd)	1.58	ND1 His76 and its symmetry mate SG Cys211 and at 2.80 Å from NZ atom of Lys66 from a symmetry related molecule SG Cys211 SG Cys211 NE2 His170 SG Cys204 ND1 His238 NE2 His238	Abe, S., Atsumi, K., Yamashita, K., Hirata, K., Mori, H., Ueno, T. (2018) Phys. Chem. Chem. Phys., **20**:2986-2989.
5YHB	Polyhedra – del(Gly192-Ala194)	Pd(allyl) complex (C_3H_5Pd)	2.08	NE2 His238 ND1 His238 NE2 His183 NE2 His223 ND1 His223 NH1 Arg209 NH2 Arg209 SG Cys211	
5E2D	Ferritin	IrCp-Pd(allyl) complex (C_3H_5Pd)	1.87	SG Cys126 NE2 His114 OE1 Glu45 OE2 Glu45 SG Cys48	Maity, B., Fukumori, K., Abe, S. Ueno, T. (2016) Chem Comm. (Camb) **52**: 5463-5466.
5HQO	Ferritin	IrCp-Pd(allyl) complex (C_3H_5Pd)	1.81	SG Cys126 NE2 His114 SG Cys48 OE2 Glu45	
5CSE	Streptadivin	Pd-containing biotin ligand	1.79	Pd is part of the biotin ligand and does not bind the protein	Chattergee, A., Mallin, H., Klehr, J., Vallapurackal, J., Finke, A.D., Vera, L., Marsh, M., Ward, T.R.

					(2016) Chem Sci - 7:673-677.
4Q9X	mTFP	$PdCl_2$	1.9	OE1 Glu123 NZ Lys102 NZ Lys32 OD2 Asp7 OE1 Glu43 OE1 Glu164 OE1 Glu117	not available.
3NOZ	Ferritin mutant E45C/R52H	Pd(allyl) complex (C_3H_5Pd)	1.52	SG Cys126 NE2 His114 ND1 His52 SG Cys48 NE2 His49 SG Cys45 NE2 His173	Wang, Z., Takezawa, Y., Aoyagi, H., Abe, S., Hikage, T., Watanabe, Y., Kitagawa, S., Ueno, T.(2011) Chem Comm., (Camb) **47** 170-172.
3NP0	Ferritin mutant E45C/H49A/ R52H	Pd(allyl) complex (C_3H_5Pd)	1.48	SG Cys 126 NE2 His114 SG Cys48 ND1 His52 SG Cys45 ND1 His173	
3NP2	Ferritin mutant E45C/C48A	Pd(allyl) complex (C_3H_5Pd)	1.86	SG Cys 126 NE2 His114 SG Cys45 NE2 His49	
3AF7	Ferritin	25Pd(allyl) complex (C_3H_5Pd)	1.58	ND1 His49 SG Cys48 SG Cys 126 NE2 His114	Abe, S., Hikage, T., Watanabe, Y., Kitagawa, S., Ueno, T. (2010) Inorg Chem 49: 6967-6973.
3AF8	Ferritin mutant C126A	Pd(allyl) complex (C_3H_5Pd)	1.66	OE1 Glu45 SG Cys48 ND1 His49	
3AF9	Ferritin mutant C48A	Pd(allyl) complex (C_3H_5Pd)	1.85	SG Cys 126 NE2 His114	
3ESC	Cut2a, cutinase-NCN-pincer	ethyl 4-nitrophenyl P-[3-(4-(bromopallado)-1,3-bis[(methylthio)methyl]-phenyl)propyl]phosphonate (C_{21} H_{27} Br N O_5 P Pd S_2)	1.2	Pd is part of the molecule and does not directly bind the protein	Rutten, L., Wieczorek, B., Mannie, J.P.B.A., Kruithof, C.A., Dijkstra, H.P., Egmond, M.R., Lutz, M., Klein Gebbink, R.J.M., Gros, P., van Koten, G.(2009) Chemistry **15**: 4270-4280.
3ESD	Cut2a, cutinase-NCN-pincer	ethyl 4-nitrophenyl P-[3-(4-(bromopallado)-1,3-bis[(methylthio)methyl]-phenyl)propyl]phosphonate (C_{21} H_{27} Br N O_5 P Pd S_2)	1.22	Pd is part of the molecule and does not directly bind the protein	
2ZG7	Ferritin mutant H49A	Pd(allyl) complex (C_3H_5Pd)	1.7	SG Cys126 NE2 His114 SG Cys48 OE2 Glu45 ND1 His49	Abe, S., Niemeyer, J., Abe, M., Takezawa, Y., Ueno, T., Hikage, T., Erker, G., Watanabe, Y. (2008) J Am Chem
2ZG8	Ferritin mutant H49A	Pd(allyl) complex (C_3H_5Pd)	1.6	SG Cys126 SG Cys48	

				OE2 Glu45 NE2 His114 OE1 Glu45	Soc **130:** 10512-10514.
2ZG9	Ferritin mutant H114A	Pd(allyl) complex (C_3H_5Pd)	1.75	SG Cys126 SG Cys48 ND1 His49	
3FI6	Ferritin mutant H49A	Pd ions	1.8	SG Cys48 OD2 Asp38 NE2 His124 NE2 His173 NE2 His114	Ueno, T., Abe, M., Hirata, K., Abe, S., Suzuki, M., Shimizu, N., Yamamoto, M., Takata, M., Watanabe, Y.(2009) J Am Chem Soc **131:** 5094-5100.
2Z5P	Ferritin	Pd ions	1.65	SG Cys126 OE1 Glu130 OE2 Glu130 SG Cys48 OE1 Glu45 NE2 His114 NE2 His49	
2Z5Q	Ferritin	Pd ions	2.1	ND1 His173 OE1 Glu45 NH2 Arg52 SG Cys126 NE2 His114 OE1 Glu53	
2Z5R	Ferritin	Pd ions	2.5	NE2 His124 SG Cys48 NE2 His114 OE1 Glu136 OE2 Glu136 NH1 Arg64 NH2 Arg64	
3BZ4	Fab22-4	Pd ions	1.8	O Asp 1 (E) N Asp 1 (E) OD1 Asp 1 (E) O Asp 1 (A) N Asp 1 (A)	Vulliez-Le Normand, B., Saul, F.A., Phalipon, A., Belot, F., Guerreiro, C., Mulard, L.A., Bentley, G.A.(2008) Proc Natl Acad Sci U S A **105:** 9976-9981.
3C6S	Fab22-4	Pd ions	1.8	N Asp 1 (A) O Asp 1 (A) N Asp 1 (E) O Asp 1 (E) OD1 Asp 1 (A)	
1KS4	Endoglucanase	Pd ions	2.5	OE1 Glu116 SD Met118 NE2 His46 N Val33 O Val33	Khademi, S., Zhang, D., Swanson, S.M., Wartenberg, A., Witte, K., Meyer, E.F. (2002) Acta Crystallogr D Biol Crystallogr. **58**:660-667.

Table S4. Structures of Lysozyme adducts with Pt compounds reported in the Protein Data Bank.

PDB code	Pt compound	Resolution (Å)	Pt-protein interaction (< 2.5 Å)	Reference
5IHG	derivative of a platin(II) compound containing a O, S bidentate ligand	1.75	ND1 His 15	Hildebrandt, J., Hafner, N., Gorls, H., Kritsch, D., Ferraro, G., Durst, M., Runnebaum, I.B., Merlino, A., Weigand, W. (2016) Dalton Trans **45**: 18876-18891.
5II3	derivative of a platin(II) compound containing a O, S bidentate ligand	1.78	ND1 His 15	
5ILC	derivative of a platin(II) compound containing a O, S bidentate ligand	1.75	ND1 His 15 (A) ND1 His 15 (B)	
5ILF	derivative of a platin(II) compound containing a O, S bidentate ligand	1.85	ND1 His 15	
5M1Y 1LKR 4OW9	Cisplatin	1.90	ND1 His 15	Helliwell, J. R. and Tanley S. W. M.. Preprint: Atomic resolution X-ray crystal structure of cisplatin bound to hen egg white lysozyme stored for 5 years 'on the shelf'. (2016) DOI: https://doi.org/10.5281/zenodo.155068
5LXW	Cisplatin 5 year on the shelf	1.0	ND1 His 15 NE2 His 15 NH1 Arg 14 NH2 Arg 14	Not available
5L3H 4DD4	Cisplatin	1.7	ND1 His 15 NE2 His 15 N Lys1 NZ Lys 96	Helliwell, J.R., Schreurs, A.M.M., Kroon-Batenburg, L.M.J., Tanley, S. (2016) ArXiv

			NZ Lys 33	
5L3I	Cisplatin	1.7	ND1 His 15 NE2 His 15 NH2 Arg 14 N Lys1 NZ Lys 96 NZ Lys 33	
5HMV	Cisplatin	0.98	ND1 His 15 NE2 His 15 N Lys1	Tanley, S.W.M., Helliwell, J.R. (2016) Struct Dyn. **3**: 037101.
5HQ1	Cisplatin	1.0	ND1 His 15 NE2 His 15	
5I5Q	Cisplatin	1.0	ND1 His 15	
5IDD	Cisplatin	1.0	ND1 His 15 NE2 His 15	
5F9U	Cisplatin	1.85	ND1 His 15 NE2 His 15 NH1 Arg 14	Ferraro, G., Pica, A., Russo Krauss, I., Pane, F., Amoresano, A., Merlino, A. (2016) J.Biol.Inorg.Chem. **21**: 433-442.
5F9X	Cisplatin	1.94	ND1 His 15 NE2 His 15 NH1 Arg 14	
5FCP	Cisplatin	1.55	ND1 His 15 NE2 His 15 NH1 Arg 14	
5HL	Cisplatin	1.7	ND1	Tanley, S.W., Schreurs, A.M.,

L			His 15 NE2 His 15 NH2 Arg 14	Kroon-Batenburg, L.M., Helliwell, J.R. (2016) Acta Crystallogr.,Sect.F **72**: 253-254.
5HMJ 4XAN	Carboplatin	1.30	ND1 His 15	Tanley, S.W., Schreurs, A.M., Kroon-Batenburg, L.M., Helliwell, J.R. (2016) Acta Crystallogr.,Sect.F **72**: 251-252.
4Z3M	derivative of a platin(II) compound containing a S, O bidentate ligand	2.15	ND1 His 15	Mugge, C., Marzo, T., Massai, L., Hildebrandt, J., Ferraro, G., Rivera-Fuentes, P., Metzler-Nolte, N., Merlino, A., Messori, L., Weigand, W. (2015) Inorg.Chem. **54**: 8560-8570.
4Z41	derivative of a platin(II) compound containing a S, O bidentate ligand	1.89	ND1 His 15	
4Z46	Cisplatin and Oxaliplatin	1.85	OD2 Asp119 ND1 His 15	Marasco, D., Messori, L., Marzo, T., Merlino, A. (2015) Dalton Trans **44**: 10392-10398.
4ZEE	Cisplatin and Oxaliplatin	1.95	OD2 Asp119 ND1 His 15	
4YEM	Carboplatin	1.47	ND1 His 15 NE2 His 15 NZ Lys 96 NH2 Arg 14	Shabalin, I., Dauter, Z., Jaskolski, M., Minor, W., Wlodawer, A. (2015) Acta Crystallogr.,Sect.D **71**: 1965-1979.
4YEN	Cisplatin	2.0	ND1 His 15 NE2 His 15 NH1 Arg 14	
4YEO	Cisplatin	0.98	NE2 His 15 NZ Lys 116 N Lys1	
4R6C	K_2PtBr_6	1.7	Non-covale	Helliwell, J.R., Brink, A., Kaenket, S., Starkey, V.L., Tanley, S.W. (2015) Faraday Discuss **177**: 429-

			ntly bound to the protein	441.
4PPO	Oxaliplatin	1.73	OD2 Asp119	Messori, L., Marzo, T., Merlino, A. (2014) Chem.Commun.(Camb.) **50**: 8360-8362.
4NSG	Carboplatin	2.0	ND1 His15 NE2 His15 NH1 Arg14	Tanley, S.W., Diederichs, K., Kroon-Batenburg, L.M., Levy, C., Schreurs, A.M., Helliwell, J.R. (2014) Acta Crystallogr F Struct Biol Commun **70**: 1135-1142.
4NSH	Carboplatin	2.1	ND1 His15 NE2 His15 NH1 Arg14	
4NSI	Carboplatin	2.3	ND1 His15 NE2 His15 NH2 Arg14	
4NSJ	Carboplatin	1.7	ND1 His15	
4OWA	Carboplatin	1.796	ND1 His15	Tanley, S.W., Helliwell, J.R. (2014) Acta Crystallogr.,Sect.F **70**: 1127-1131.
4OWB	Cisplatin	1.69	ND1 His15 NE2 His15 NH1 Arg14	
4OWC	K_2PtI_6	1.62	ND1 His15	Tanley, S.W., Starkey, L.V., Lamplough, L., Kaenket, S., Helliwell, J.R. (2014) Acta Crystallogr.,Sect.F **70**: 1132-1134.
4OWE	K_2PtCl_6	1.41	Non-covalently bound to	

			the protein	
4OWH	K_2PtBr_6	1.48	Non-covalently bound to the protein	
2XTH	K_2PtBr_6	1.8	Non-covalently bound to the protein	Helliwell, J.R., Bell, A.M.T., Bryant, P., Fisher, S., Habash, J., Helliwell, M., Margiolaki, I., Kaenket, S., Watier, Y., Wright, J., Yalamanchili, S.K. (2010) Z.Kristallogr. **225**: 570
3WPK	potassium hexachloroplatinate	2.0	Pt^{2+} at 4.3 Å from ND1 atom of Asn65 Pt^{2+} at 4.6 Å from NH1 Arg14	Mizutani, R., Shimizu, Y., Saiga, R., Ueno, G., Nakamura, Y., Takeuchi, A., Uesugi, K., Suzuki, Y. (2014) Sci Rep **4**: 5731-5731.
3WPL	potassium hexachloroplatinate	2.0	NE2 His15 (to be filled) OD1 Asn65 Pt^{2+} at 3.0 Å from OXT Leu129	
3WU7	potassium hexachloroplatinate	2.0	Pt^{2+} at 4.3 Å from ND1 atom	

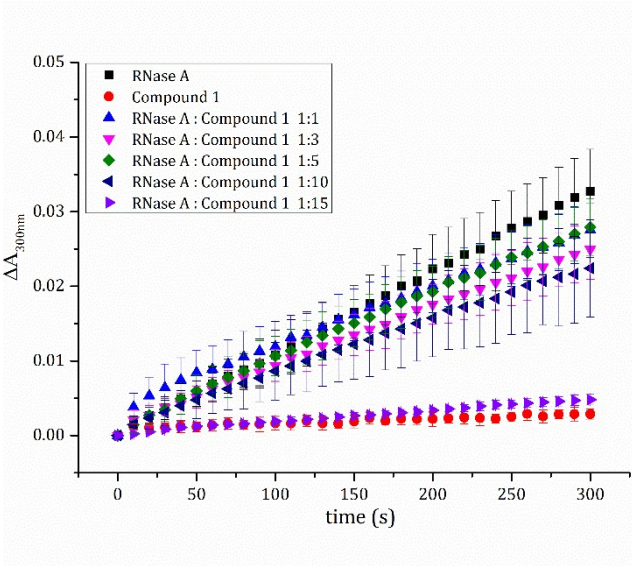
			of Asn65 Pt^{2+} at 4.6 Å from NH1 Arg14	
3WU8	potassium hexachloroplatinate	2.0	2.7 Å from NE2 His15 (chain to be flipped) and 2.6 Å from NH1 Arg14	
3WU9	potassium hexachloroplatinate	2.0	Pt^{2+} at 4.6 Å from ND1 atom of Asn65 Pt^{2+} at 4.6 Å from NH1 Arg14 Pt^{2+} at 4.7 Å from OXT Leu129	
3WUA	potassium hexachloroplatinate	2.0	NE2 His15 (chain to	

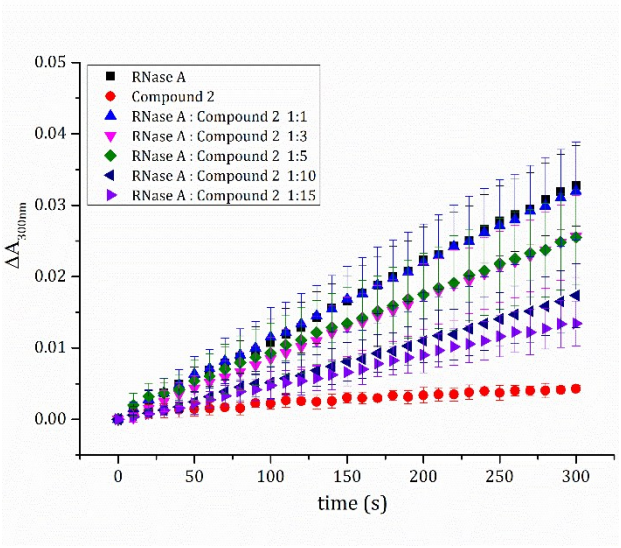
			be flipped) and 2.8 Å from NH1 Arg 14 Pt^{2+} at 2.7 Å from OD1 Asn65 Pt^{2+} at 4.7 Å from OXT Leu129	
4QGZ	trans-dimethylamine methylamine dichlorido platinum(II)	2.51	NE2 His 15 OD2 Asp101	Messori, L., Marzo, T., Michelucci, E., Russo Krauss, I., Navarro-Ranninger, C., Quiroga, A.G., Merlino, A. (2014) Inorg.Chem. **53**: 7806-7808.
4LT0	Carboplatin	2.1	ND1 His 15	Tanley, S.W., Diederichs, K., Kroon-Batenburg, L.M., Levy, C., Schreurs, A.M., Helliwell, J.R. (2014) Acta Crystallogr.,Sect.F **70**: 1135-1142.
4LT3	Carboplatin	2.0	ND1 His 15	
4LT1	Carboplatin	2.3	ND1 His 15	Tanley, S., Diederichs, K., Kroon-Batenburg, L.M.J.. M. M Schreurs, Antoine & Helliwell, J. (2013). Carboplatin binding to a model protein in non-NaCl conditions to eliminate partial conversion to cisplatin, and the use of different criteria to choose the resolution limit. AirXs. 2013arXiv1309.4661T
4LT2	Carboplatin	2.0	NH1 Arg 14	
4MR1	cis-diamminediiodoplatinum(II)	1.99	ND1 His 15	Messori, L., Marzo, T., Gabbiani, C., Valdes, A.A., Quiroga, A.G., Merlino, A. (2013) Inorg.Chem. 52: 13827-13829
3TXF	Cisplatin	1.69	ND1 His 15	Tanley, S.W., Schreurs, A.M., Helliwell, J.R., Kroon-Batenburg, L.M. (2013) J.Appl.Crystallogr. **46**: 108-119.
3TXG	Cisplatin	1.7	ND1 His 15 NE2	

			His 15	
3TXH	Cisplatin	1.69	ND1 His 15 NE2 His 15	
3TXI	Cisplatin	1.6	ND1 His 15 NE2 His 15 NH1 Arg 14	
3TXK	Cisplatin	3.0	ND1 His 15 NE2 His 15	
4GCB	Cisplatin and Carboplatin	1.8	ND1 His 15 NE2 His 15 NH2 Arg 14	Helliwell, J.R., Tanley, S.W. (2013) Acta Crystallogr.,Sect.D **69**: 121-125.
4GCC	Cisplatin and Carboplatin	2.0	ND1 His 15 NE2 His 15 NH1 Arg 14	
4GCD	Cisplatin and Carboplatin	2.8	ND1 His 15 NE2 His 15	
4GCE	Cisplatin and Carboplatin	2.9	ND1 His 15 NE2 His 15	
4GCF	Cisplatin and Carboplatin	3.5	ND1 His 15 NE2 His 15	
4G49	Cisplatin	2.4	ND1 His 15 NE2 His 15	Tanley, S.W., Schreurs, A.M., Kroon-Batenburg, L.M., Helliwell, J.R. (2012) Acta Crystallogr.,Sect.F **68**: 1300-1306.
4G4B	Cisplatin	2.1	ND1 His 15 NE2	

			His 15 NH1 Arg 14	
4G4C	Carboplatin	2.0	ND1 His 15 NE2 His 15	
4G4H	Carboplatin	2.0	ND1 His 15 NE2 His 15	
3AZ5	CoPt nanoparticles	2.34	NE2 His 15 N Lys1	Abe, S., Tsujimoto, M., Yoneda, K., Ohba, M., Hikage, T., Takano, M., Kitagawa, S., Ueno, T. (2012) Small **8**: 1314-1319.
3AZ7	CoPt nanoparticles	2.1	ND1 His 15 NE2 His 15	
4DD7	Carboplatin	1.6	ND1 His 15 NE2 His 15 NH1 Arg 14	Tanley, S.W., Schreurs, A.M., Kroon-Batenburg, L.M., Meredith, J., Prendergast, R., Walsh, D., Bryant, P., Levy, C., Helliwell, J.R. (2012) Acta Crystallogr.,Sect.D **68**: 601-612.
4DD9	Carboplatin	1.6	ND1 His 15 NE2 His 15 NH1 Arg 14	
4DDB	Cisplatin	3.0	ND1 His 15 NE2 His 15	
4DDC	Cisplatin	1.8	ND1 His 15 (A) NE2 His 15 (A) NE2 His 15 (B)	
2DQA	K_2PtCl_4	1.6	SD Met29 (A) SD Me	Goto, T., Abe, Y., Kakuta, Y., Takeshita, K., Imoto, T., Ueda, T. (2007) J.Biol.Chem. **282**: 27459-27467.

			t29 (B)	
2I6Z	Cisplatin	1.9	ND1 His 15	Casini, A., Mastrobuoni, G., Temperini, C., Gabbiani, C., Francese, S., Moneti, G., Supuran, C.T., Scozzafava, A., Messori, L. (2007) Chem.Comm.(Camb.) --: **156**-158.





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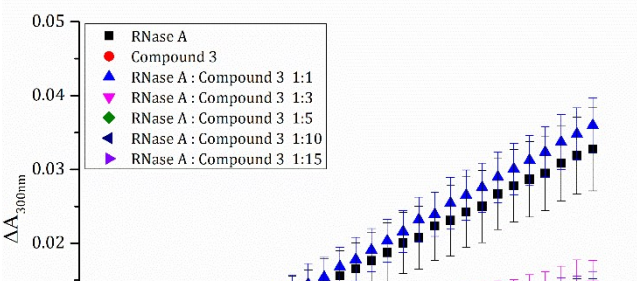

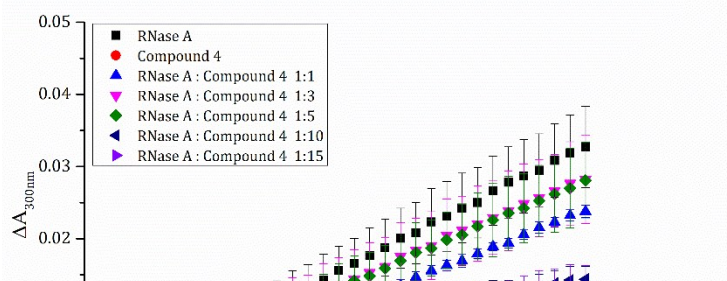

Figure S1. Ribonuclease activity of RNase A and of the investigated RNase A-metal compound adducts. The enzymatic activity on yeast RNA has been evaluated by measuring the variation of absorbance at 300 nm as function of time upon addition of the protein to the yeast RNA sample. Results for RNase A are reported in black, whereas those for the free compounds used as controls are in red. Experiments have been performed using the Kunitz method[1] in 0.05 M sodium acetate buffer at pH 5.5 at different protein to metal molar ratios.

[1]M. Kunitz, *Journal of Biological Chemistry* 1946, **164**, 563−568.